Notice of Allowability	Application No.	Applicant(s)	
	09/646,925	CHATFIELD, STEVEN NEVILLE	
	Examiner	Art Unit	
	Vanessa L. Ford	1645	

-- The MAILING DATE of this communication appears on the cover sheet with the correspondence address--

All claims being allowable, PROSECUTION ON THE MERITS IS (OR REMAINS) CLOSED in this application. If not included herewith (or previously mailed), a Notice of Allowance (PTOL-85) or other appropriate communication will be mailed in due course. **THIS NOTICE OF ALLOWABILITY IS NOT A GRANT OF PATENT RIGHTS.** This application is subject to withdrawal from issue at the initiative of the Office or upon petition by the applicant. See 37 CFR 1.313 and MPEP 1308.

1. ☒ This communication is responsive to *30 January 2004*.

2. ☒ The allowed claim(s) is/are *1, 5-11 and 16-17*.

3. ☒ The drawings filed on *25 September 2000* are accepted by the Examiner.

4. ☒ Acknowledgment is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d) or (f).
   a) ☒ All b) ☐ Some* c) ☐ None of the:
      1. ☒ Certified copies of the priority documents have been received.
      2. ☐ Certified copies of the priority documents have been received in Application No. _____ .
      3. ☐ Copies of the certified copies of the priority documents have been received in this national stage application from the International Bureau (PCT Rule 17.2(a)).

   * Certified copies not received: _____.

Applicant has THREE MONTHS FROM THE "MAILING DATE" of this communication to file a reply complying with the requirements noted below. Failure to timely comply will result in ABANDONMENT of this application.
THIS THREE-MONTH PERIOD IS NOT EXTENDABLE.

5. ☐ A SUBSTITUTE OATH OR DECLARATION must be submitted. Note the attached EXAMINER'S AMENDMENT or NOTICE OF INFORMAL PATENT APPLICATION (PTO-152) which gives reason(s) why the oath or declaration is deficient.

6. ☐ CORRECTED DRAWINGS (as "replacement sheets") must be submitted.
   (a) ☐ including changes required by the Notice of Draftsperson's Patent Drawing Review (PTO-948) attached
      1) ☐ hereto or 2) ☐ to Paper No./Mail Date _____.
   (b) ☐ including changes required by the attached Examiner's Amendment / Comment or in the Office action of Paper No./Mail Date _____.

   Identifying indicia such as the application number (see 37 CFR 1.84(c)) should be written on the drawings in the front (not the back) of each sheet. Replacement sheet(s) should be labeled as such in the header according to 37 CFR 1.121(d).

7. ☐ DEPOSIT OF and/or INFORMATION about the deposit of BIOLOGICAL MATERIAL must be submitted. Note the attached Examiner's comment regarding REQUIREMENT FOR THE DEPOSIT OF BIOLOGICAL MATERIAL.

Attachment(s)

1. ☐ Notice of References Cited (PTO-892)
2. ☐ Notice of Draftperson's Patent Drawing Review (PTO-948)
3. ☒ Information Disclosure Statements (PTO-1449 or PTO/SB/08), Paper No./Mail Date 10/20/2003
4. ☐ Examiner's Comment Regarding Requirement for Deposit of Biological Material
5. ☐ Notice of Informal Patent Application (PTO-152)
6. ☒ Interview Summary (PTO-413), Paper No./Mail Date *7/26/2004* .
7. ☒ Examiner's Amendment/Comment
8. ☒ Examiner's Statement of Reasons for Allowance
9. ☐ Other _____.

EXAMINER'S AMENDMENT

1. An examiner's amendment to the record appears below. Should the changes and/or additions be unacceptable to applicant, an amendment may be filed as provided by 37 CFR 1.312. To ensure consideration of such an amendment, it MUST be submitted no later than the payment of the issue fee.

Authorization for this examiner's amendment was given in a telephone interview with Gary Tanigawa on July 26, 2004.

2. This Office Action is responsive to Applicant's response January 30, 2004. All rejections of record are withdrawn in view of Applicant's amendments and remarks.

3. Claims 1, 5-11 and 17 are directed to an allowable product. Pursuant to the procedures set forth in the Official Gazette notice dated March 26, 1996 (1184 O.G. 86), claim 16, directed to the process of using the patentable product, previously withdrawn from consideration as a result of a restriction requirement, directed to a method of raising an immune response in a mammalian host is now subject to being rejoined. Process claim 16 is hereby rejoined and fully examined for patentability under 37 CFR 1.104. Therefore claims 1, 5-11, 16 and 17 are allowed an renumbered 1-10 respectively.

4. The application has been amended as follows:

In the specification:

Page 11, replace the first paragraph starting with line 1 with:

Formulation of the vaccine

The vaccine may be formulated using known techniques for formulating attenuated bacterial vaccines. The vaccine advantageously presented for oral administration, example lyophilised encapsulated form. Such capsules may be provided with an enteric coating comprising, example, EUDRAGIT ~~Eudragate~~"S" (Trade Mark) anionic polymer of methacrylic acid and methacrylates with a –COOH group, EUDRAGIT ~~Eudragate~~ "L" (Trade Mark) anionic polymer of methacrylic acid and methacrylates with a –COOH group, cellulose acetate, cellulose phthalate hydroxypropylmethyl cellulose. These capsules may be used as such, alternatively, the lyophilised material may be reconstituted prior to administration, as suspension. Reconstitution advantageously effected in a buffer at a suitable pH to ensure the viability the bacteria. In order to protect the attenuated bacteria and the vaccine from gastric acidity, a sodium bicarbonate preparation advantageously administered before each administration of the vaccine. Alternatively, the vaccine may be prepared for parenteral administration, intranasal 20 administration or intramuscular administration.

Page 21, replace the seventh paragraph starting on line 35 with:

Cells from the grown plate were streaked onto the following media and grown overnight at 37°C [[37oc]].

Page 24, replace the second paragraph starting on line 28 with:

6) Southern blotting of PTL003:

Structure of deletion mutations: Total DNA was extracted from cultures of the three deletion mutants grown form the microbanked stocks, digested with restriction endonuclease EcoRV and the digested DNA was subjected to pulsed filed agarose gel electrophoresis. DNA was blotted from the gels onto HYBOND ~~Hybond~~ N+ (Trade Name) nylon membranes and hybridised with appropriate DNA probes according to standard procedures. Results (Figure 6) show that the hybridizing chromosomal DNA fragments of the mutants are shorter than the wild-type, consistent with the mutations being deletions.

Page 25, replace the second paragraph starting on line 7 with:

Confirmation of absence of Heat-Stable (ST) and Heat-Labile (LT) toxin genes in E. coli strain E1392/75-2A. For this the ST and LT-AB genes were used as DNA probes against total DNA from E1392/75-2A. Total DNA from the toxin positive ETEC strain E1393/75 was included as a positive control, while that from the laboratory E. coli strain JM109 was included as a negative. Hybridised membranes were left under HYPERFILM-ECL ~~Hyperfilm ECL~~ (Trade Name) emulsion for 1 h to obtain the

maximum amount of signal. Probes were prepared using PCR with plasmid DNA extracted from E1392/75-2A as template and oligonucleotides EST01 and EST02 as primer for ST, or LT-R1 and LT-03 for LT-AB. There was no significant hybridization with total DNA using either the LT-AB or ST probe, despite obtaining a very intense signal from the positive control total DNA.

In the claims:

A. Claim 1. (previously presented) An *Escherichia coli* bacterium attenuated by a non-reverting mutation in each of the *aroC* gene, the *ompF* gene and the *ompC* gene.

B. Claims 2-4 (cancelled).

C. Claim 5. (previously presented) An *Escherichia coli* bacterium according to claim 1 which is a strain of enterotoxigenic *E. coli* (ETEC).

D. Claim 6. (previously presented) An *Escherichia coli* bacterium according to claim 1 which is further attenuated by a mutation in a fourth gene.

E. Claim 7. (amended) An *Escherichia coli* bacterium according to claim 6 wherein the fourth gene is selected form the group consisting of *aroA, AroD, aroE ,~~aroS,~~ pur, htrA, galE, cya, crp, phoP and surA.*

F. Claim 8. (amended) An *Escherichia coli* bacterium according to claim 1, wherein the mutation in each and every gene is a defined mutation.

G. Claim 9. (amended) An *Escherichia coli* bacterium according to claim 1, wherein the mutation in each and every gene is deletion of the entire coding sequence.

H. Claim 10. (previously presented) An *Escherichia coli* bacterium according to claim 1 which has been genetically engineered to express a heterologous antigen.

I. Claim 11. (amended) An *Escherichia coli* bacterium according to claim 1, wherein expression of the antigen is driven by the *nirB* ~~*nilrB*~~ promoter or the *htrA* promoter.

J. Claims 12 –15 (cancelled)

K. Claim 16. (original) A method of raising an immune response in an mammalian host, which comprises administering to the host an *Escherichia coli* bacterium as defined in claim 1.

L. Claim 17. (previously presented) An *Escherichia coli* bacterium according to claim 1, which is PTL003 deposited on September 3, 2001 under accession number 01090302 with the European Collection of Cell Cultures (ECACC).

5. The following is an examiner's statement of reasons for allowance. The prior art cited neither teaches nor suggests an *Escherichia coli* bacterium attenuated by a non-reverting mutation in each of the aroC gene, the ompF gene and the ompC gene. Nor does that prior art teach a method of raising an immune response in a mammalian host comprising administering to the host an *Escherichia coli* bacterium attenuated by a non-reverting mutation in each of the aroC gene, the ompF gene and the ompC gene.

Any comments considered necessary by applicant must be submitted no later than the payment of the issue fee and, to avoid processing delays, should preferably accompany the issue fee. Such submissions should be clearly labeled "Comments on Statement of Reasons for Allowance."

Conclusion

6. Any inquiry of the general nature or relating to the status of this general application should be directed to the Group receptionist whose telephone number is (703) 308–0196.

Papers relating to this application may be submitted to Technology Center 1600, Group 1640 by facsimile transmission. The faxing of such papers must conform with the notice published in the Office Gazette, 1096 OG 30 (November 15, 1989). Should applicant wish to FAX a response, the current FAX number for the Group 1600 is (703) 872-9306.

Any inquiry concerning this communication from the examiner should be directed to Vanessa L. Ford, whose telephone number is (571) 272-0857. The examiner can normally be reached on Monday – Friday from 9:00 AM to 6:00 PM. If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, Lynette Smith, can be reached at (571) 272-0864.

Information regarding the status of an application may be obtained from the Patent Application Information Retrieval (PAIR) system. Status information for published applications may be obtained from either Private PAIR or Public PAIR. Status information for unpublished applications is available through Private PAIR only. For more information about the PAIR system, see <http://pair-direct.uspto.gov./>. Should you have questions on access to the Private PAIR system, contact the Electronic Business Center (EBC) at 866-217-9197 (toll-free).

Vanessa L. Ford
Biotechnology Patent Examiner
August 3, 2004

NITA MINNIFIELD
PRIMARY EXAMINER
8/5/04

CLEAN COPY OF CLAIMS

1. An *Escherichia coli* bacterium attenuated by a non-reverting mutation in each of the *aroC* gene, the *ompF* gene and the *ompC* gene.

5. An *Escherichia coli* bacterium according to claim 1 which is a strain of enterotoxigenic *E. coli* (ETEC).

6. An *Escherichia coli* bacterium according to claim 1 which is further attenuated by a mutation in a fourth gene.

7. An *Escherichia coli* bacterium according to claim 6 wherein the fourth gene is selected from the group consisting of *aroA, aroD, aroE, pur, htrA, galE, cya, crp, phoP and surA.*

8. An *Escherichia coli* bacterium according to claim 1, wherein the mutation in each and every gene is a defined mutation.

9. An *Escherichia coli* bacterium according to claim 1, wherein the mutation in each and every gene is deletion of the entire coding sequence.

10. An *Escherichia coli* bacterium according to claim 1 which has been genetically engineered to express a heterologous antigen.

11. An *Escherichia coli* bacterium according to claim 1, wherein expression of the antigen is driven by the *nirB* promoter or the *htrA* promoter.

16. A method of raising an immune response in an mammalian host, which comprises administering to the host an *Escherichia coli* bacterium as defined in claim 1.

17. An *Escherichia coli* bacterium according to claim 1, which is PTL003 deposited on September 3, 2001 under accession number 01090302 with the European Collection of Cell Cultures (ECACC).